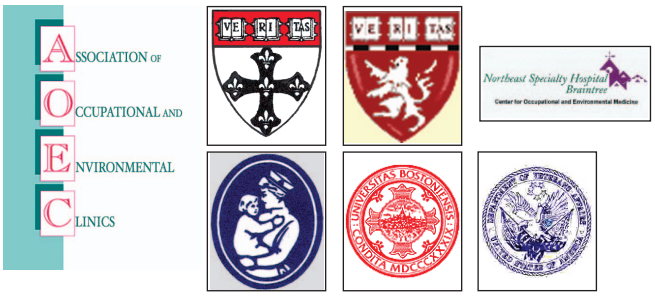
Cognitive deficits and magnetic resonance spectroscopy in adult monozygotic twins with lead poisoning.



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Cognitive Deficits and Magnetic Resonance Spectroscopy in Adult Monozygotic Twins with Lead Poisoning

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Seventy-one-year-old identical twin brothers with chronic lead poisoning were identified from an occupational medicine clinic roster. Both were retired painters, but one brother (J.G.) primarily removed paint and had a history of higher chronic lead exposure. Patella and tibia bone lead concentrations measured by K-X-ray fluorescence in each brother were 5–10 times those of the general population and about 2.5 times higher in J.G. than in his brother (E.G.). Magnetic resonance spectroscopy (MRS) studies examined *N*-acetylaspartate:creatine ratios, a marker of neuronal density. Ratios were lower in J.G. than in his brother. Scores on neurocognitive tests that assess working memory/executive function were below expectation in both twins. Short-term memory function was dramatically worse in J.G. than in his brother. These results demonstrate some of the more subtle long-term neurologic effects of chronic lead poisoning in adults. In particular, they suggest the presence of frontal lobe dysfunction in both twins, but more dramatic hippocampal dysfunction in the brother with higher lead exposure. The MRS findings are consistent with the hypothesis that chronic lead exposure caused neuronal loss, which may contribute to the impairment in cognitive function. Although a causal relation cannot be inferred, the brothers were genetically identical, with similar life experiences. Although these results are promising, further study is necessary to determine whether MRS findings correlate both with markers of lead exposure and tests of cognitive function. Nevertheless, the results point to the potential utility of MRS in determining mechanisms of neurotoxicity not only for lead but also for other neurotoxicants as well.
Key words: **lead poisoning, magnetic resonance spectroscopy, monozygotic, neuropsychological tests, paint, twins.** ***Environ Health Perspect*** **112:620–625 (2004). doi:10.1289/ehp.6687 available via *http://dx.doi.org/* [Online 8 January 2004]**

Case Presentation

J.G. and E.G. are 71-year-old monozygotic twins. Both are retired painters who worked in the Boston metropolitan area. The brothers worked together but performed different, well-defined tasks: J.G. removed paint by scraping, sanding, and heat treatment with an electric iron; E.G. predominantly painted but at times assisted in paint removal. J.G. smoked cigarettes and ate at work without washing his hands. E.G. smoked cigarettes until the early 1990s but reported that he was meticulous about washing his hands before eating at work. Both wore paper masks at work but did not use any sophisticated respiratory protective devices.

In 1984, J.G. developed chronic back pain, for which he was referred to a neurosurgeon. Because of his occupational history of painting, a blood lead (BPb) level was ordered and returned at 125 μg/dL. He was subsequently hospitalized and chelated with ethylenediamine tetra-acetic acid (EDTA). Since his chelation he has been followed in the Center for Occupational and Environmental Medicine at what is now the Northeast Specialty Hospital. J.G.'s other chronic health problems include hypertension, which E.G. has as well. Otherwise both brothers are healthy. Both E.G. and J.G. graduated from the same high school, served together in the Navy, and have worked together as painters for > 45 years. The combination of differential lead exposures in the context of complete genetic matching and similar childhood and adult environments provided a unique opportunity for assessment of central nervous system (CNS) effects of lead. Cognitive testing and magnetic resonance spectroscopy (MRS) studies were employed to determine differences that could be attributed to differential lead exposures.

Neurocognitive testing. Both brothers were tested on a battery of cognitive tests, including the Wechsler Adult Intelligence Scale-Revised (WAIS-R) (Wechsler 1981) and the Mini-Mental State Exam (MMSE) (Folstein et al. 1975). A battery of specific tests designed to assess the domains of attention, executive function, verbal and language skills, visuospatial abilities, manual motor speed and dexterity, memory and behavior/personality was also performed.

J.G. underwent cognitive testing in both 1990 and 1999. Based on his reported academic history and his performance on tests of verbal and academic ability that are relatively impervious to the effects of CNS insults in adulthood, his premorbid verbal/language abilities were judged to be at the lower end of the average range and his visuospatial skills to be at the upper end of the average range. In

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We are indebted to the steadfast and patient cooperation of J.G. and E.G., the two study participants who were the focus of this investigation.

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1990, his performance was below expectation for estimated premorbid abilities in the domains of manual motor skills, attention, working memory/executive function, and visuospatial abilities. Assessment of short-term memory function showed deficits at the level of learning new information on several tasks, but his retention of newly learned information over delays was normal (i.e., he did not show significant forgetting of information over delays). In 1999, J.G.'s performance was within expected limits for estimated premorbid abilities in the domain of attention; however, he performed below expectation in the domains of motor function, working memory/executive function, and visuospatial functioning. On testing of short-term memory, performance was below expectation at the levels of both learning and retention of newly learned information (i.e., he showed significant forgetting). When comparing his 1999 performance with that in 1990, we used age-adjusted outcome measures to control for the age increase. His simple attention appeared to improve. The most dramatic decline was seen in the area of short-term memory, although his manual motor control was also somewhat worse. Scores on tasks assessing the domains of working memory/executive function and his drawings remained below expectation.

E.G. was tested only in 1999. Like his brother, he consistently did better on visuospatial than on verbal tasks, with verbal/language skills at the lower end of the average range and visuospatial skills at the upper end of the average range. Test performance was below expectation for estimated premorbid abilities in the domains of motor skills, working memory/executive function, and visuospatial abilities. On short-term memory tests, he performed somewhat below expectation at the level of learning on two tasks, but his retention of newly learned information over delays was normal.

A comparison of the 1999 assessments of each twin showed that both had mild manual motor deficits, but these appeared to be more pronounced in J.G. Scores on tests assessing working memory/executive function were likewise below expectation for both twins, although on slightly different tasks. Short-term memory function was dramatically worse in J.G. than in E.G., involving both the processes of learning and retention. The neuropsychological test results are shown in Table 1. Several test scores within each domain were judged by the neuropsychologist to be abnormal on the basis of estimated premorbid skills for each brother. The cognitive test results are consistent with frontal lobe dysfunction in both twins, but with rather dramatic hippocampal dysfunction in J.G.

Bone lead levels and MRS. In 1998, bone lead measurements were taken with a K-X-ray fluorescence (KXRF) bone lead analyzer (Aro et al. 1994; Chettle et al. 1991). We determined the ratio of *N*-acetylaspartate (NAA) to creatine, a marker of neuronal density, using MRS. From each brother we obtained 1.5-tesla single-voxel point resolved spectroscopy (PRESS) spectra [repetition time ÷ echo time = 2,000/144 msec/msec with 128 averages] from five voxels. The voxel locations were in the left and right frontal lobes, the left and right hippocampi, and one voxel in the left midbrain encompassing the central semiovale and selected from the same axial slice as the frontal lobe voxels. Voxel sizes were roughly 1.7 cm^3. Spectral analysis was performed with software supplied by the manufacturer (SA/GE; General Electric Medical Systems, Milwaukee, WI) and consisted of spectral phasing followed by peak area fitting for the metabolite resonances of choline, creatine, and NAA. Independent spectral analyses were performed by two individuals experienced with daily processing of clinical single voxel spectra, and the results from each region and operator were averaged. Examples of spectra from the left frontal lobes of each twin are shown in Figure 1. The bone lead concentrations and NAA:creatine ratios from the MRS exams are summarized in Table 2. J.G. had much higher levels of trabecular (patella) lead and cortical (tibia) lead than did E.G. In general, J.G. demonstrated a decrease of 10–30% in the NAA:creatine ratio compared with E.G.

Discussion

The case of these twins illustrates many of the classic clinical and public health issues

Table 1. Neuropsychological test[a] results by functional domain.

Tests organized by functional domain	J.G. 1990	J.G. 1999	E.G. 1999
General intelligence			
WAIS-R			
Verbal IQ[b]	94	98	93
Performance IQ[b]	101	111	103
Fullscale IQ[b]	96	103	96
MMSE	28/30	27/30	27/30
Wide Range Achievement Test–III			
Reading[b]	NA	94	104
Attention/executive function (expected = average)			
WMS-R			
Attention index[b]	85[c]	95	96
Digit span forward	18th percentile[c]	46th percentile	46th percentile
Digit span backward	72nd percentile	66th percentile	66th percentile
Visual spans forward	1st percentile[c]	44th percentile	44th percentile
Visual spans backward	48th percentile	62nd percentile	62nd percentile
Mental control	6/6	4/6[c]	5/6
WAIS-R			
Digit spans age-scaled scores[d]	9	9	9
Arithmetic age-scaled scores[d]	9	7[c]	8
Trail-Making Test A	0 errors	0 errors	0 errors
Trail-Making Test B	3 errors[c]	1 errors	2 errors[c]
Continuous Performance Test	NA	13 errors[c]	19 errors[c]
Verbal, language (expected = low average)			
WAIS-R			
Information age-scaled scores[d]	10	12	12
Vocabulary age-scaled scores[d]	8	9	9
Similarities age-scaled score[d]	10	8	8
Comprehension[d]	10	13	9
Controlled word association	25–29th percentile	25–29th percentile	< 10th percentile[c]
Boston Naming Test	40/60	43/60	50/60
Visuospatial/visual motor (expected = average)			
Finger tapping test (avg no. taps in 5 trials of 10 sec each)			
Right	74.2 (dom)	65.2 (dom)	58
Left	61.4	41.8[c]	53 (dom)
Grooved pegboard (seconds to place all pegs, avg of 2 trials)			
Right	NA	83 (dom)	83
Left	NA	88	88 (dom)
WAIS-R			
Picture completion[d]	9	11	10
Digit symbol[d]	8	10	11
Picture arrangement[d]	11	11	12
Block design[d]	16	13	10
Object assembly[d]	11	14	12
Boston Visuospatial Quantitative Battery drawings	Impaired[c] Perseverative[c]	Small[c] Impaired[c] Perseverative[c]	Impaired[c] Perseverative[c]
Rey-Osterrieth figure copy	NA	> 16th percentile	75–99th percentile

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surrounding acute and chronic adult lead toxicity and offers a unique opportunity to explore the utility of MRS for examining effects of lead exposure. Although both twins had elevated body burdens of lead, there were differences between them, which, in identical twins with very similar life exposures, provided an ideal opportunity to explore the use of MRS technology for assessing the impact of lead toxicity on the CNS and perhaps shed light on mechanisms of action.

Lead exposure in Massachusetts. Construction work has become the dominant source of lead exposure for adults in the United States. In Massachusetts, 1 of the 27 states that currently maintain central registries of blood lead tests and report surveillance data to the National Institute for Occupational Health and Safety and Health Adult Blood Lead Epidemiology and Surveillance Program [Centers for Disease Control and Prevention (CDC) 1999], construction workers accounted for 63% of 381 individuals identified with BPb levels of ≥ 40 µg/dL—the action level in the Occupational Health and Safety Administration's (OSHA) standard (Rabin et al. 1994). Most houses in the United States built before 1978 (estimated at 42–47 million houses) have lead-based paint inside and outside [Agency for Toxic Substances and Disease Registry (ATSDR) 1998]. Lead paint can contain up to 50% lead by weight, which poses an enormous risk to construction workers—including painters—who remove it as well as to children whose hand-to-mouth behavior and frequent floor activity raise their risk of ingesting lead paint chips and lead-contaminated house dust. Scraping, and, in particular, sanding lead paint creates a fine lead dust that can be easily inhaled. Absorption of lead is highly efficient after inhalation, particularly if the particles are small. Hand-to-mouth behavior of construction workers can also lead to significant absorption of lead, such as smoking cigarettes and eating without prior hand washing. Lead dust on the hands can be ingested and absorbed through the gastrointestinal tract as can lead dust on cigarettes, which can be heated during smoking generating lead fumes that are especially well absorbed by the lungs. In addition to use in residences, lead paint was also used in commercial buildings and other structures, such as bridges. Workers who remove paint in these sectors are at extremely high risk for lead exposure (Levin and Goldberg 2000). Construction work is regulated under the OSHA construction lead standard that took effect in 1993 (OSHA 1993), and some states have additional standards that apply specifically to the painting and deleading of residences. Such regulations require the use of certain personal protective equipment (e.g., special respirators) and work techniques that reduce exposure (e.g., "wet scraping" to reduce dust), as well as prohibit certain activities that increase exposure (e.g., smoking and eating at work). These regulations, however, are often difficult to enforce and do not apply to individual homeowners who undertake renovations themselves.

Neurocognitive effects of lead. Although lead has adverse effects on numerous health end points (ATSDR 1999), the most sensitive target of lead exposure is the nervous system. Neurologic functions for which there is evidence of an adverse effect of chronic exposure to lead include peripheral nerve conduction velocity, postural balance, visual and auditory evoked potentials, cardiac autonomic nervous system function, and neurocognitive functions mediated by the CNS (Araki et al. 2000;

Table 1. Continued

Tests organized by functional domain	J.G. 1990	J.G. 1999	E.G. 1999
Short-term memory (expected = average)			
WMS-R indices			
General memory[b]	99	97	100
Verbal memory[b]	93	93	89
Visual memory[b]	111	107	115
Delayed recall[b]	111	90[c]	111
WMS-R			
Orientation	14/14	14/14	NA
Logical memory			
Immediate recall	44th percentile	28th percentile	24th percentile
Delayed recall	37th percentile	37th percentile	46th percentile
Paired associate learning			
Immediate recall	15/24[c]	16/24[c]	16/24[c]
Delayed recall	8/8	5/8[c]	8/8
Difficult paired associate learning			
Immediate recall	13/40[c]	6/40[c]	4/40[c]
Delayed recall	5/40[c]	2/40[c]	2/40[c]
Consonant trigrams	31/60[c]	NA	NA
California Verbal Learning Test			
Trials 1–5[e]	NA	–4[c]	0
Interference list[e]	NA	–1[c]	0
Short delay recall[e]	NA	–3[c]	–1[c]
Short delay cued recall[e]	NA	–3[c]	0
Long delay free recall[e]	NA	–5[c]	+1
Long delay cued recall[e]	NA	–5[c]	0
Recognition[d]	NA	–5[c]	+1
WMS-R			
Figural memory	5/10[c]	6/10[c]	6/10[c]
Visual paired associate learning			
Immediate recall	13/18[c]	12/18[c]	7/18[c]
Delayed recall	5/6	6/6	6/6
Visual reproduction			
Immediate recall	98th percentile	52th percentile	98th percentile
Delayed recall	98th percentile	56th percentile	94th percentile
Mood/personality			
Profile of Mood States	Confusion[c] Fatigue[c]	Fatigue[c]	WNL
Minnesota Multiphasic Personality Inventory	Normal profile	Increase in depression but WNL	WNL

Abbreviations: avg, average; dom, dominant hand; NA, not available; WMS-R, Wechsler Memory Scale–Revised; WNL, within normal limits.
[a]See Lezak (1998) and Spreen and Strauss (1998) for further details on individual tests. [b]Mean ± SD, 100 ± 15. [c]Below expectation based on estimated premorbid abilities. [d]Mean ± SD, 10 ± 3. [e]SDs below average.

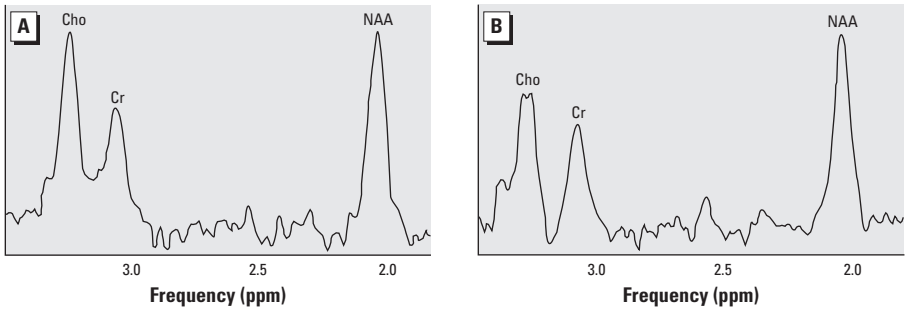

Figure 1. Spectra from the left frontal lobes of (*A*) J.G. and (*B*) E.G. Abbreviations: Cho, choline; Cr, creatine.

Balbus-Kornfeld et al. 1995; Cheng et al. 1998). Acute lead exposure can lead to clinical manifestations of lead poisoning in the adult, including seizures, coma, and even death, although BPb levels must be quite high (ATSDR 1999); this has become rare in the past 15–20 years. In children, such effects may be seen at lower levels [e.g., > 70–80 μg/dL (ATSDR 1999)]. Less severe neurologic and behavioral effects have been documented in lead-exposed workers with BPb levels between 40 and 120 μg/dL. Evidence consistently indicates that lead-exposed workers perform worse on tests of visual motor functioning, reaction time, memory, attention, and concentration, with effects on mood also often being noted (Arnvig et al. 1980; Baker et al. 1984; Campara et al. 1984; Grandjean et al. 1978; Haenninen et al. 1978; Hogstedt et al. 1983; Schwartz et al. 2001; Stollery 1996; Stollery et al. 1991; Valciukas et al. 1978).

The association between low-level lead exposure and neurocognitive function has been most extensively studied in children, particularly in relation to measures of IQ (Banks et al. 1997; Needleman and Gatsonis 1990; Schwartz 1994). Although the current BPb level of concern set forth by the CDC is 10 μg/dL, there may be no lower limit of BPb level at which these effects occur (Canfield et al. 2003; Schwartz 1994). Similar effects may also occur in adults at levels well below 40 μg/dL, because impairments in neurocognitive functioning in several domains have recently been associated with very low BPb levels (Muldoon et al. 1996; Payton et al. 1998). However, this issue remains unresolved (Seeber et al. 2002). The half-life of lead in blood is only about 1 month, so BPb levels may reflect only relatively recent exposure. To the extent that bone lead acts as a source of lead in blood, however, BPb can reflect longer-term exposure.

In a review of the evidence that cumulative exposure to lead impairs cognitive function in adults, Balbus-Kornfeld et al. (1995) concluded that the evidence was not strong. The authors acknowledged that their conclusions, however, may possibly have more to do with the availability of good measures of cumulative exposure than a lack of a true association (Balbus-Kornfeld et al. 1995). Since that time, several other studies have assessed this association. Three studies using multiple BPb measurements to create an integrated measure of cumulative exposure among occupationally exposed populations reported cross-sectional associations of this measure with lower neurobehavioral test scores (Chia et al. 1997; Lindgren et al. 1996; Lucchini et al. 2000), although one such study failed to find an association with their cumulative lead measure (Barth et al. 2002). Several other studies used KXRF technology to assess cumulative lead exposure (the half-life of lead in bone is decades) and its association with cognitive performance. Most of these studies were cross-sectional in design and involved occupationally exposed populations. One of these with a small sample size (n = 57) did not find a relation between bone lead and neurobehavioral tests (Osterberg et al. 1997) and two others found small effects of bone lead (Hanninen et al. 1998; Schwartz et al. 2001), whereas the others reported more robust associations between higher bone lead and worse neurocognitive performance (Bleecker et al. 1997; Fiedler et al. 2003; Stewart et al. 1999). Two other studies of an elderly general (nonoccupational) population of men found that higher bone lead was associated with impairments on neuropsychological tests of visual memory and spatial copying (Payton et al. 1998) and increased odds of scoring < 24 on the MMSE—a traditional cut point for increased risk of dementia (Wright et al. 2003). In addition, studies have suggested that both the *ApoE* genotype and education level interact with the cumulative effect of lead on cognitive performance (Bleecker et al. 2002; Stewart et al. 2002). The only study to examine longitudinal decline in cognitive function found that higher tibia lead levels predicted declines in verbal memory and learning, visual memory, executive ability, and manual dexterity among former lead workers (Schwartz et al. 2000).

Table 2. Summary of bone lead and BPb levels and NAA:creatine ratios.

	J.G.	E.G.
Bone lead		
Patella, 1998 (μg/g bone)[a]	343 ± 9.4	119 ± 8.8
Tibia, 1998 (μg/g bone)[a]	189 ± 7.8	79 ± 7.2
BPb (μg/dL)		
November 1989	88	33
October 1990	51	20
Fall 1994[b]	51	15
May 1998	30	ND
NAA:creatine ratio[c]		
Hippocampus	1.30 ± 0.10	1.60 ± 0.20
Frontal lobe	1.15 ± 0.17	1.52 ± 0.48
Midbrain	1.47 ± 0.02	1.65 ± 0.02

ND, no data.
[a]Measured value ± uncertainty. [b]November for J.G. and September for E.G. [c]Mean ± SD.

The pattern of cognitive deficits in the twins that we report here is generally quite typical of the pattern of deficits reported after high-level lead exposure. This pattern includes predominant impairments in the domains of attention/executive function, visuospatial/visual motor functioning, short-term memory, and (for J.G.) confusion and fatigue, whereas verbal language and general intelligence remain relatively unimpaired. Test of single-word reading, basic written arithmetic, and semantic knowledge (e.g., the ability to name common objects) are not generally sensitive to exposure to neurotoxicants in adults. Disruptions of these types of cognitive functions are usually seen only after widespread brain damage (e.g., frank hypoxia, severe traumatic brain injury, Alzheimer disease after the initial stages) or focal strokes involving highly specific brain areas that mediate language and calculations. For these reasons, neuropsychologists often use these tests when evaluating adults with suspected CNS insults to estimate premorbid patterns and levels of cognitive function in different domains (especially verbal, visuospatial, and attention). After exposure to toxicants such as lead in adulthood, cognitive deficits tend to be specific, not generalized and not affecting language centers in the brain. In the case of lead, this is probably due to its action on hippocampal and frontal areas of the brain. In a recent study of cumulative (bone lead) exposure in a general population, Wright et al. (2003) found a significant association with slightly lower scores on the MMSE. Overall, J.G. scored lower on the neurocognitive testing than did his brother (E.G.), which is consistent with J.G.'s higher bone lead levels and lower NAA:creatine ratios. In the case of the twins presented here, however, we cannot distinguish what effects might be related to high acute BPb concentrations as opposed to cumulative exposure reflected in the high bone lead levels. It should also be noted that other known neurotoxicants such as solvents are frequently used in painting. Some of the functional deficits noted in this study may in part be related to toxicants other than lead, and differential exposure to these other neurotoxicants could also contribute to some of the differences on cognitive tests between the twins.

The magnetic resonance images (MRIs) from the twins showed lesions indicative of microinfarcts. This is consistent with known adverse effects of lead on the cardiovascular system. In the context of the neurobehavioral deficits exhibited by the twins, it is possible that these outcomes are to some extent the result of adverse cerebrovascular events brought about as the result of chronic lead exposure. Such effects would constitute an indirect action of lead on neuronal density and neurobehavioral impairment through actions on the

cerebrovascular system, in addition to the likely direct effects on the nervous system.

MRS measurements of neuronal density. The effects of elevated blood and bone lead levels have been examined primarily in the context of behavioral and neuropsychologic evaluations. There has been a growing interest in the mechanisms by which lead disrupts brain function. Although the adverse effects of lead exposure on neurobehavioral functioning is one of the most consistently reported impairments associated with lead exposure, little is known about the effects of lead on brain metabolism *in vivo* or about the structural and functional correlates of lead-related brain dysfunction.

MRS provides a noninvasive method with which to monitor biochemical aspects of acute and chronic stages of neurologic disease in the human brain. The development of spatially localized spectroscopic methods that sample the relative levels of metabolites from volumes of tissue defined from MRI scans has provided a basis for integrating the biochemical information obtained by MRS with the anatomical and pathological information obtained from MRIs. MRS has gained widespread acceptance as a method for assessing both neuronal viability and demyelination. MRS can detect both NAA and creatine in discrete tissue volumes. In the cortex, NAA is located in neuronal cell bodies, whereas in the white matter, it is located largely in axons. A decrease in NAA has been proposed as an indicator of neuronal and axonal damage and loss (Arnold and De Stefano 1997; van der Knaap et al. 1992). In practice, the decrease in NAA is measured relative to the level of creatine, a stable metabolite whose level is constant after neuronal loss.

The use of MRS to examine the effects of lead exposure is new. A report on MRS findings in a 10-year-old boy with elevated BPb levels and his 9-year-old male cousin who did not have elevated BPb showed that the lead-exposed boy had lower NAA:creatine ratios in both frontal gray and white matter (Trope et al. 1998). A subsequent study of 16 children with elevated BPb levels and 5 children whose measured BPb levels had never been > 10 μg/dL found that the children with elevated BPb levels had statistically significantly lower NAA:creatine ratios in frontal gray matter. NAA:creatine ratios were also lower for these children in frontal white matter, but this did not reach statistical significance (Trope et al. 2001). These results, as well as evidence showing reduced NAA in disease processes involving intellectual deterioration, led us to hypothesize a decrease in NAA in the brains of adults with clinical evidence of lead exposure.

In the twins presented here, J.G. had NAA:creatine ratios that were lower than those of E.G., suggesting lower neuronal density. This is consistent with the results of the neurocognitive tests, on which J.G. performed worse in general than did E.G. In addition, J.G. demonstrated declines between 1990 and 1999 on tests assessing short-term memory and visuospatial performance, suggesting a new progressive process that may be related to his history of lead exposure. Both twins, however, showed significant impairments on several neuropsychological tests, which is consistent with the fact that they both had bone lead levels that were high for their age, because both patella and tibia lead levels are typically < 40 μg/g bone in community-exposed individuals around 71 years of age (Hu et al. 1996). This also may suggest that although E.G.'s NAA:creatine ratios were higher than those of J.G., the NAA:creatine ratios seen in E.G. may be lower than expected for age-matched non-lead–exposed individuals.

The NAA:creatine ratio has been reported in a number of MRS studies of brain metabolites in control populations and populations with specific diseases such as amyotrophic lateral sclerosis (ALS) and Alzheimer disease (Barker et al. 2000; Chan et al. 1999; Doraiswamy et al. 1998; Kreis et al. 1993; Lundbom et al. 1999). It is important to note that the NAA:creatine ratio has not only regional (Barker et al. 2000; Jayasundar and Raghunathan 1997; Kreis et al. 1993; Lundbom et al. 1999; Ricci et al. 2000) and developmental dependencies but also depends on the specific echo time and repetition time of the MRS pulse sequence used to acquire the data as the relaxation times, T_1 and T_2, because the different metabolites are not identical (Kreis et al. 1993). For instance, Chan et al. (1999) reported NAA:creatine ratios in the motor cortex of 3.08 ± 0.32 for 14 healthy subjects (mean age, 57 ± 11 years) compared with 2.40 ± 0.42 for 11 patients with ALS. Lundbom et al. (1999) studied the NAA:creatine ratio in the context of the normal aging process and reported NAA:creatine ratios from 1.88 to 2.59 for five elderly volunteers (mean age, 74 ± 7) compared with 2.07 to 3.54 for seven younger volunteers (mean age 35 ± 6 years). Barker et al. (2000) measured NAA:creatine in six healthy volunteers (mean age, 38 ± 3 years) and reported values ranging from 1.75 to 3.03 depending on the brain region examined. In these three studies, repetition time values identical or comparable with our value of 2,000 msec were used. However, these studies used longer echo time values of 272 to 280 msec compared with the 144 msec used in the present study. Thus, to compare ratios, correction factors accounting for the differential T_2 decay must be applied. Assuming a mixture of gray and white matter, representative NAA and creatine T_2 values may be estimated from the study of Kreis et al. (1993) to be 441 msec and 207 msec, respectively. Thus, to compare NAA:creatine ratios from the studies mentioned above with our NAA:creatine ratios, a correction factor of approximately 0.72 must be applied. Taking the range reported by Lundbom et al. (1999) of 1.88 to 2.59 for elderly volunteers, we would expect a range of 1.35 to 1.86 for the NAA:creatine ratios at the 144-msec echo time used in our study. The range of NAA:creatine values we found in the two elderly brothers in our study were from 1.15 to 1.89, within the range if somewhat lower than what may be anticipated from normal elderly volunteers. It is important to note that the average NAA:creatine values of 1.31 ± 0.16 and of 1.59 ± 0.07 for J.G. and E.G., respectively, fall within the range anticipated from the control values estimated above. Furthermore, these values are within the range of NAA:creatine values measured by Doraiswamy et al. (1998), who used MRS sequence parameters directly comparable with ours in their study of 12 elderly (mean age, 73 ± 9 years) probable Alzheimer patients. J.G. presents a mean NAA:creatine value similar to the lowest value reported in that study. Clearly, as the field of MRS matures and NAA:creatine ratios for larger control populations at various ages are measured, more meaningful interpretations of NAA:creatine values in given individuals may be made. Within the context of the present study, although there may not be an exact relation between the NAA:creatine ratios we obtained in the twin brothers and those found in other studies, the difference in NAA:creatine ratio between the twins spans a range on the order of that seen for elderly adults in other studies.

Although we cannot conclusively attribute the differences in NAA:creatine ratios between the brothers to the differences in lead exposure, the fact that these two brothers matched for genetics, education level, and many life experiences would support the hypothesis that the lower NAA:creatine ratios in J.G. are secondary to higher lead exposure. If so, this suggests that chronic lead exposure caused a loss of neurons in the hippocampus, frontal cortex, and midbrain. Possible mechanisms of cell loss include lead-induced oxidative toxicity (Adonaylo and Oteiza 1999), cellular apoptosis without necrosis (Fox et al. 1998), and indirect oxidative toxicity via increases in the metabolite aminolevulinic acid (Bechara 1996). Clearly the study of the relation between lead exposure and neuronal density as assessed by MRS in a larger population will be necessary to determine these relations with more certainty.

Conclusions

Construction work has become the dominant source of lead exposure in U.S. adults. Neurobehavioral sequelae of lead toxicity are not uncommon and studies are beginning to suggest that these outcomes can occur with

chronic exposure at levels allowed under current U.S. regulation. Presenting symptoms of acute lead toxicity are often vague and may likely involve health end points other than neurobehavioral ones, such as the back pain that initially brought J.G.'s lead exposure to medical attention. Thus, particularly when dealing with construction workers, a high index of suspicion and a low threshold for testing BPb levels are called for in order to diagnose lead toxicity.

In the cases presented, both of the monozygotic twin painters clearly had extremely high bone lead levels. Nonetheless, their differential lead exposure resulting from different job tasks was reflected in differences in bone lead levels. On the background of genetic identity and extremely similar life exposures, the relations between lead levels, neuropsychological testing, and MRS results are highly suggestive. The markedly higher bone lead levels in J.G. were paralleled by greater deficits in neuropsychological testing performance and lower NAA:creatine ratios in the hippocampus and frontal lobes. These results are consistent with neuronal loss secondary to lead exposure, which could be responsible in part for the impaired neuropsychological function on hippocampal and frontal-lobe–dependent tasks. Although we cannot establish cause and effect, we believe that MRS may be a valuable research tool in determining the mechanisms of neurotoxicity of lead and potentially other neurotoxicants as well.

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